ERA CoBioTech (ERA-Net Cofund on Biotechnologies)

ACHEMA2018

Kick-off session: "Biotechnology for a sustainable bioeconomy"

Project name: Synthetic Biology for Industrial Production of Steroids

Project acronym: Syntheroids

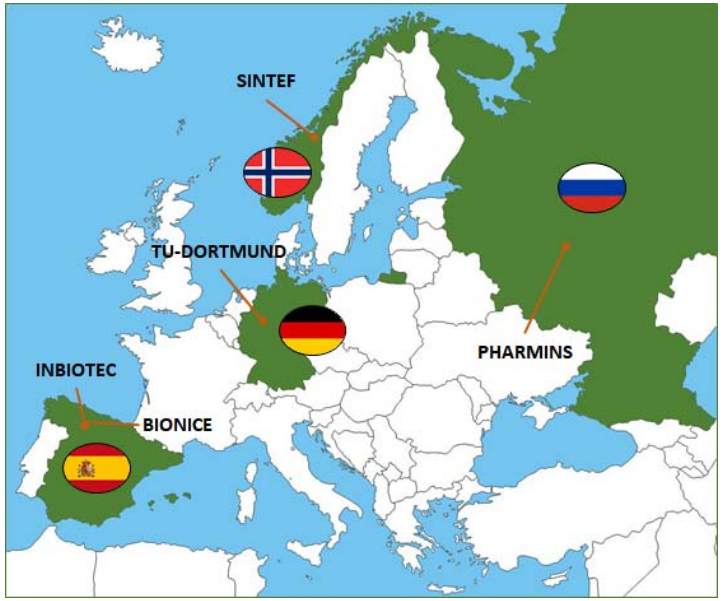
Name: Alberto Sola-Landa (INBIOTEC)

This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant 722361

Frankfurt am Main, 14.06.2018

Project partners

- INBIOTEC (research institute), Spain
- Pharmins Ltd. (company), Russia
- SINTEF, (research institute) Norway
- TU- Dortmund (university), Germany
- BioNice (company), Spain



- Total project budget: 2,308,000 €

- Project start: May 15th, 2018

Kick-off meeting
May 24th, 2018
León, Spain

Project partners

- ***Partner 1, Coordinator***

Dr. Alberto Sola
INBIOTEC (Institute for Biotechnology)
León, Spain
E-mail: alberto.sola@inbiotec.com

- ***Tasks in the project***
  - ✓ *Coordination and management*
  - ✓ *Genome sequencing*
  - ✓ *Transcriptomics and Proteomics*
  - ✓ *Genetic engineering*

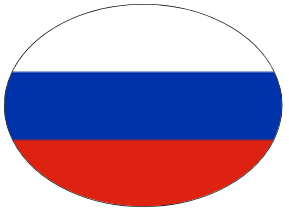
Project partners

- ***Partner 2***

Dr. Marina V. Donova

Pharmins Ltd.

Pushchino, Russia



PHARMINS
PHARMACEUTICAL INGREDIENTS

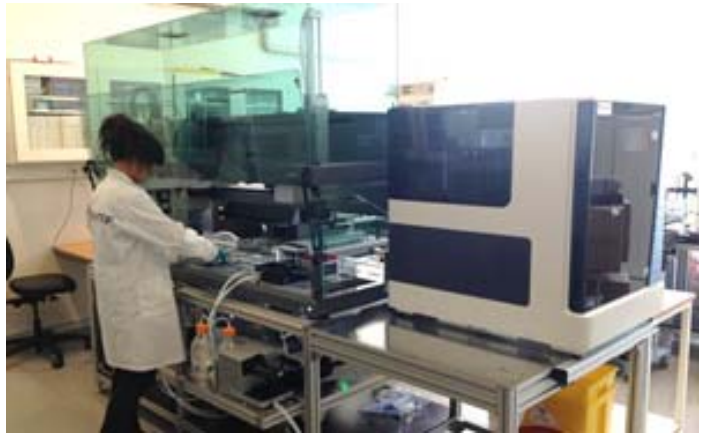
- ***Tasks in the project***
  - ✓ *Transcriptomics*
  - ✓ *Analysis of metabolic capacities*
  - ✓ *Genetic engineering*

Project partners

- ***Partner 3***

Dr. Simone Balzer Le
SINTEF Industry, Biotechnology and Nanomedicine
SINTEF AS
Trondheim, Norway

- ***Tasks in the project***
  - ✓ *Metabolomics*
  - ✓ *Genetic engineering*
  - ✓ *Process design for bioproduction*
  - ✓ *Coordination of DM activities*



Project partners

Partner 4

Dr.-Ing Gerhard Schembecker
Laboratory of plant and process design
Technical University of Dortmund
Dortmund, Germany

technische universität dortmund

Tasks in the project

- ✓ *Downstream processes*
- ✓ *Economic evaluation of process alternatives*
- ✓ *Integrate up and downstream processing*
- ✓ *Elaborate the LCA*

Project partners

Partner 5

Dr. José Luis Barredo
Department of Biotechnology
Bionice S.L.U.
León, Spain

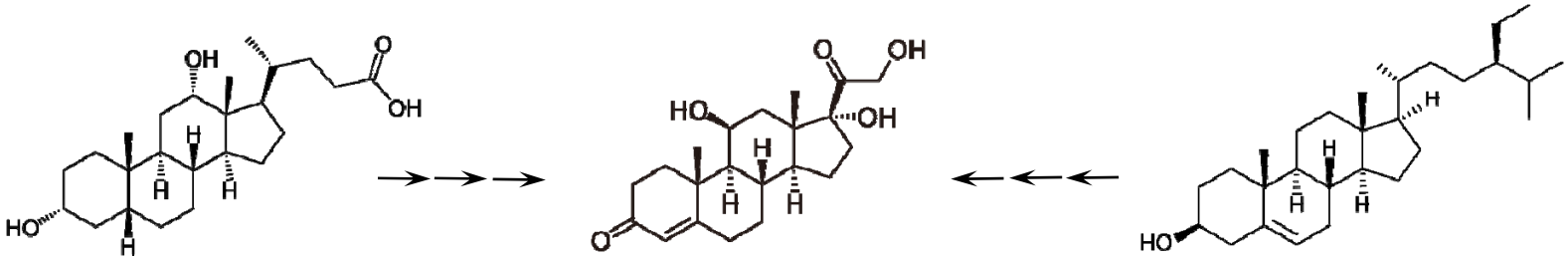
Tasks in the project

- ✓ *Analysis of steroid production*
- ✓ *Fermentation development*
- ✓ *Demonstration at industrial scale*

Steroids

- Second largest class of drugs
- More than 300 clinically approved steroidal compounds
- Clinical use since 1949
- Multiple applications
  - ✓ Rheumatoid arthritis
  - ✓ Neurodegenative diseases
  - ✓ Cancer
  - ✓ Inflammatory diseases
  - ✓ Metabolic disorders
  - ✓ Contraception
  - ✓ Hormonal insufficiencies
  - ✓ Others

Use of microorganisms for steroid production



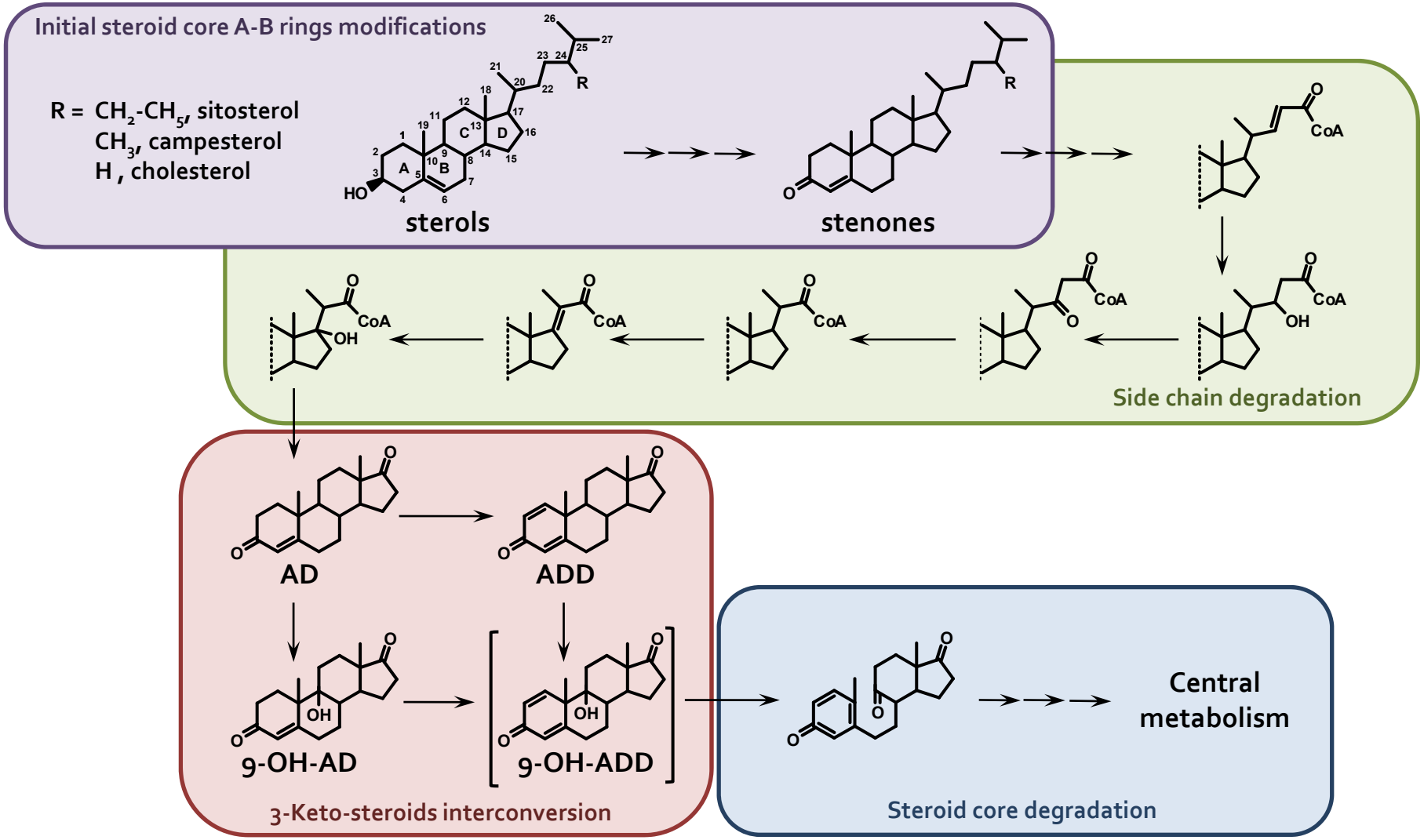
Deoxycholic acid → **Cortisone** ← **Sitosterol**

✓ **1949**
- Chemical synthesis
- Deoxycholic acid
- 31 steps
- $200 per gram

✓ **1952**
- Fungal C11 hydroxylation
- Deoxycholic acid
- 11 steps
- $6 per gram

✓ **1980**
- Mycobacterium mutants
- Phytosterol
- $0.46 per gram

Introduction



Introduction

*The **central objective** of Syntheroids is **to develop an integrated production process for pharmaceutical steroids** using Synthetic Biology and improved processing technology. Syntheroids project has four **specific objectives**:*

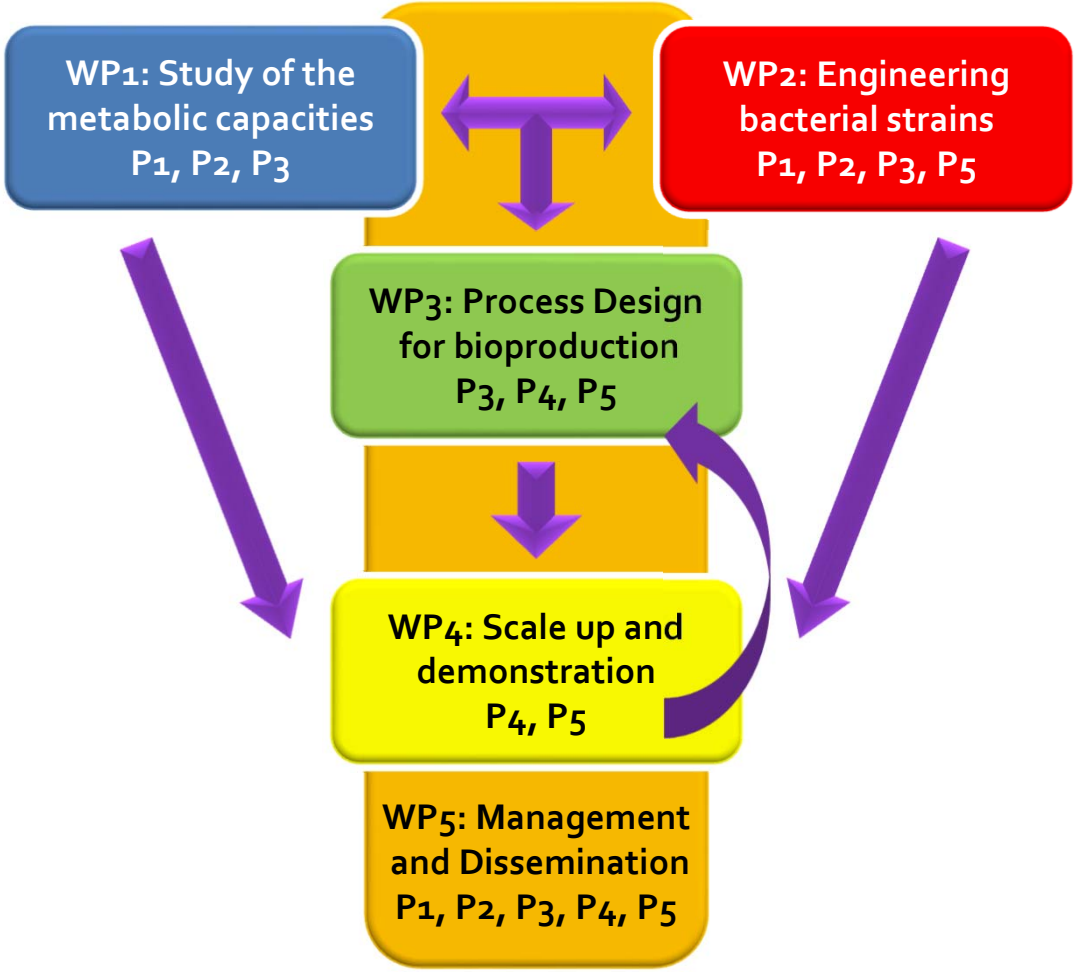
- ***Omics data integration** from steroid producing Actinobacteria as a source of Synthetic Biology targets for productive strain evolution.*
- ***Creating genetically engineered bacterial strains** capable of producing innovative **C22-steroid precursors**.*
- ***Reduce or eliminate end-product inhibition** by mutagenesis, genetic engineering and process optimization.*
- ***Integrate up- and downstream processes** for an eco-friendly bioconversion.*

Introduction

- ***Scientific approaches***
  - ✓ *Synthetic biology*
  - ✓ *Use of bioinformatics*
  - ✓ *Biotechnological approaches*

- ***Project topic areas***
  - ✓ *Sustainable production and **conversion of** different types of feedstocks and **bioresources into added value products***
  - ✓ *Development of new products, **value-added products** and supply services*
  - ✓ ***Sustainable industrial processes***

Project plan

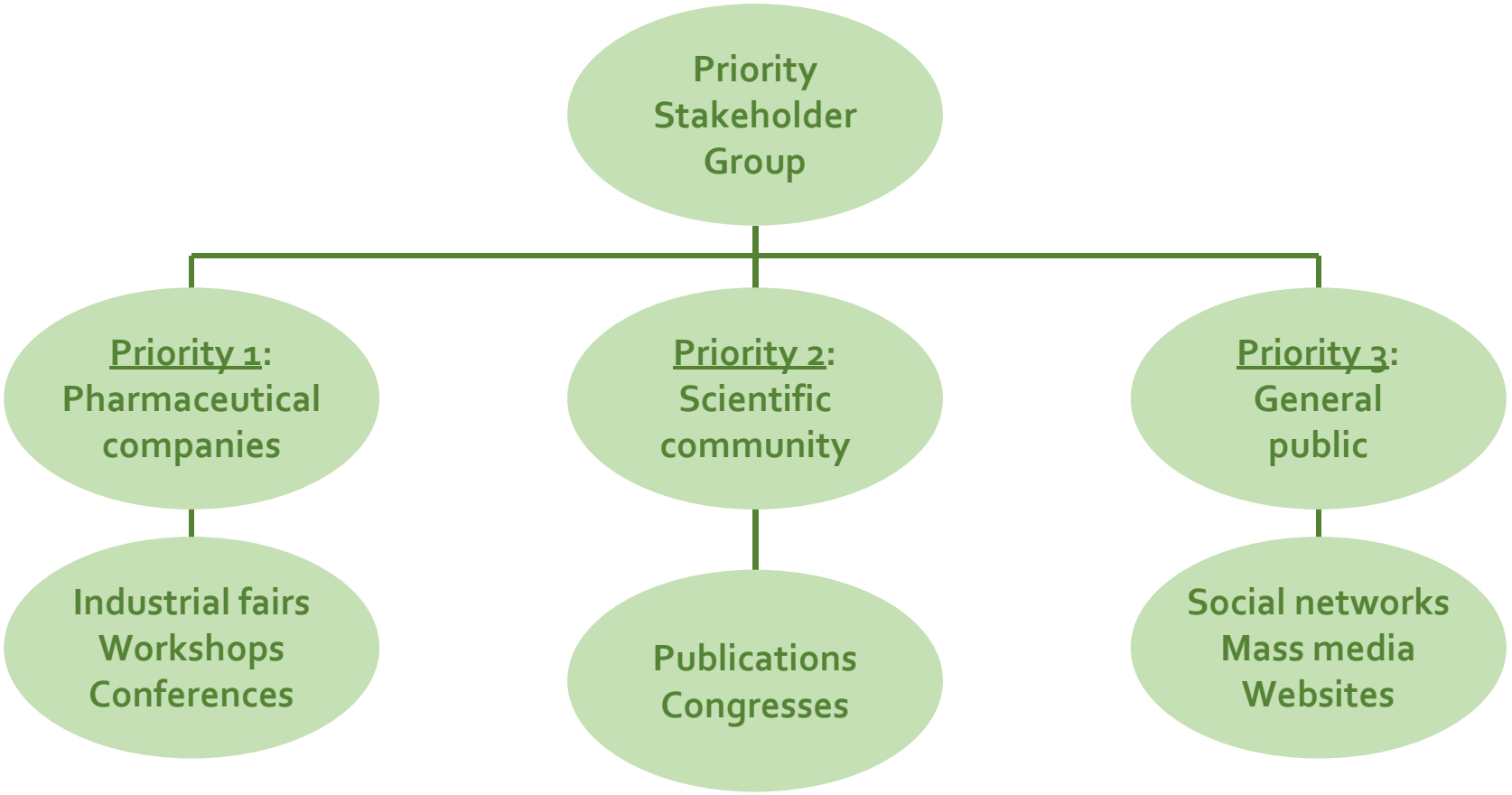


Project plan

- ***Data Management***
  - ✓ *Collaboration with and subcontracting to FAIRDOM*
  - ✓ *All raw data stored at each partner, local hub for data sharing (at SINTEF) , final data migrated into online hub (FAIRDOM Hub)*
  - ✓ *Data generated:*
    - Genome sequencing
    - Transcriptomics
    - Proteomics
    - Metabolomics
    - LC/GC-MS analysis
    - Fermentation
    - Simulation/Modelling
    - Downstream processing

Project plan

- *Communication Plan*



Project plan

- ***Life Cycle Assessment***
  - ✓ *Robot based **conceptual design methodology***
    - *Robot based HTS for miniaturized and automated experiments*
    - *Key performance indicators*
    - *Modelling to monitor and predict experimental conditions*
  - ✓ *Identification of **cost drivers** for the unit operations and for fermentation*
  - ✓ *Rating different **feeding strategies** and **process evaluation** in early stages*
  - ✓ *Inclusion of **mass and energy balances***
  - ✓ *Inclusion of **recycle and waste streams***
  - ✓ *LCA from "**cradle-to-factory gate**"*

Expected outcomes

- ***Outcomes to be achieved***

  - ✓ *To increase the **product portfolio** and decrease **end-product inhibition** combining Synthetic Biology and 'omics analyses*

  - ✓ *To ease **product purification** using a robot-based conceptual design methodology together with modelling and simulation*

  - ✓ *To decrease production costs with a better **integration of industrial procedures** (up- to downstream)*

Expected outcomes

Planed implementation and exploitation of results

- ✓ *Bionice will **validate** the steroid production processes in a **relevant industrial environment** using strict approaches to standardize, scale-up and monitor the manufacturing, according to GLP and GMP standards*

- ✓ *Bionice will provide to Crystal Pharma **improved fermentation and purification processes** for starting materials enabling the production increase, including some new compounds not produced today*

- ✓ *Pharmins will create a **platform for effective new bioprocesses** for value-added steroids from phytosterols, widening the portfolio of microbiotechnologies of the company*

Summary

- ***What is proposed***
  - ✓ *To generate effective engineered strains*
  - ✓ *To improve bioprocess efficiency*
  - ✓ *To improve product recovery*

- ***What should be achieved***
  - ✓ *A wider spectrum of bio-based steroids*
  - ✓ *A decreased by-product production*
  - ✓ *Reduced end-product inhibition*
  - ✓ *A microbial chassis for efficient and selective eco-friendly phytosterol bioconversion*